



An investigation to enhance understanding of the stimulation of weed seedling emergence by soil disturbance

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Summary

Enhanced understanding of soil disturbance effects on weed seedling recruitment will help guide improved management approaches. Field experiments were conducted at 16 site-years at 10 research farms across Europe and North America to (i) quantify superficial soil disturbance (SSD) effects on *Chenopodium album* emergence and (ii) clarify adaptive emergence behaviour in frequently disturbed environments. Each site-year contained factorial combinations of two seed populations (local and common, with the common population studied at all site-years) and six SSD timings [0, 50, 100, 150, 200 day-degrees (d°C, base temperature 3°C) after first emergence from undisturbed soil]. Analytical units in this study were emergence flushes. Flush magnitudes (maximum weekly emergence per count flush) and flush frequencies (flushes year⁻¹) were compared between disturbed and undisturbed seedbanks. One year after burial, SSD promoted seedling emergence relative to undisturbed seedbanks by increasing flush magnitude rather than

increasing flush frequency. Two years after burial, SSD promoted emergence through increased flush magnitude and flush frequency. The promotional effects of SSD on emergence were strongest within 500 d°C following SSD; however, low levels of SSD-induced emergence were detected as late as 3000 d°C following SSD. Accordingly, stale seedbed practices that eliminate weed seedlings should occur within 500 d°C of disturbance, because few seedlings emerge after this time. However, implementation of stale seedbed practices will probably cause slight increases in weed population densities throughout the year. Compared with the common population, local populations exhibited reduced variance in total emergence measured within sites and across SSD treatments, suggesting that *C. album* adaptation to local pedo-climatic conditions involves increased consistency in SSD-induced emergence.

Keywords: tillage, seedbank management, stale seedbed, genotype–environment interactions, seed germination, *Chenopodium album*.

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Introduction

Pre-plant seedbed preparation and other forms of disturbance confined to the top 10 cm of soil [hereafter referred to as ‘superficial soil disturbances’ (SSD)] can increase weed seedling emergence compared with undisturbed soil (Egley, 1989). SSD promotion of emergence is the foundation for management interventions that deplete weed seedbanks by stimulating germination and eliminating subsequent seedlings with non-selective control (e.g. stale seedbed tactics) (Riemens *et al.*, 2007). SSD-induced weed emergence may be inevitable in crop production, as suggested by Longchamps *et al.* (2012), who determined that disturbances as small as wheel tracking could promote weed seedling emergence. Promotion of weed seedling emergence by SSD remains difficult to predict due to microclimate effects, population–environment interactions and farming technique–environment interactions that have impacts on the timing and magnitude of emergence events following SSD.

Combined results from previous studies indicate that: (i) SSD promotion of seedling emergence occurs by increasing the density of a given recruitment cohort (hereafter referred to as ‘flush’) rather than increasing flush frequency (Roberts & Potter, 1980; Ogg & Dawson, 1984; Myers *et al.*, 2005), and (ii) the stimulatory effect of a particular SSD event dissipates over time, such that flushes occurring long after the SSD event feature seedling densities similar to concurrent flushes from undisturbed soil (Roberts & Potter, 1980; Ogg & Dawson, 1984; Mulugeta & Stoltenberg, 1997; Chauhan *et al.*, 2006). These observations may represent a general framework for understanding SSD-induced emergence. However, the foundational studies were each characterised by the absence of probability calculations, one seed population per species, and few (one or two) site-years of data collection. Thus, current knowledge of emergence dynamics following SSD are primarily based on visual comparisons of emergence phenologies and are therefore potentially limited in scope. Here, we strengthen general knowledge of SSD-induced emergence, by using multiple seed populations

collected and buried across a broad geographic scale to quantify population and environmental effects on SSD-induced emergence.

Regeneration from seeds can involve distinct processes of physiological dormancy loss, germination and pre-emergence seedling growth (Benech-Arnold *et al.*, 2000). Dormancy is an internal seed characteristic that prevents the completion of germination in environments otherwise suitable for plant growth (Vleeshouwers *et al.*, 1995). As dormancy decreases in response to specific combinations of temperature and moisture conditions integrated over time, germination is promoted by exposure to species-specific environmental cues including, but not limited to: light, fluctuating and constant temperatures, and gaseous germination stimulants. These environmental cues for the late steps in dormancy termination (Finch-Savage & Leubner-Metzger, 2006), alternatively defined as triggers of germination (Vleeshouwers *et al.*, 1995), are potentially provided by SSD. Once germination is completed (as indicated by radicle protrusion), pre-emergence seedling growth proceeds in response to soil temperature and soil moisture. Promotion of seedling emergence by SSD is indirect; by providing triggers for germination, SSD modifies seeds to respond to environmental drivers for emergence (temperature and moisture). SSD-induced emergence is thus subject to site-year variation in the burial environment (Mulugeta & Stoltenberg, 1997; Myers *et al.*, 2005).

Emergence behaviour can differ between seed populations, reflecting differences in dormancy loss and germination caused by: parental genotypes, maternal inheritance of plastids, maternal contributions to embryo-covering structures and maternal provisioning during seed development, which is subject to environmental constraints (Donohue & Schmitt, 1998). Population effects can cause seed populations to respond dissimilarly to a common stimulus and necessitate different models and approaches for management of weed populations in specific areas (Clements *et al.*, 2004). Although populations are known to influence many aspects of regeneration from seeds, population effects

have yet to be studied with respect to SSD-induced emergence.

In agricultural fields, SSD events are not strong predictors of environments conducive to seedling recruitment because subsequent disturbances may terminate newly emerged seedlings prior to lifecycle completion. In response to the low fidelity between germination cues and recruitment potential, plants have evolved to produce seed populations comprising individuals with quantitative and qualitative differences in germination requirements (Childs *et al.*, 2010). This ensures that only a portion of the total population germinates following a particular SSD event. Regulated sensitivity to germination cues spreads emergence across multiple germination events occurring over weeks, months and years, which reduces variance in fitness in temporally variable environments (Childs *et al.*, 2010). Accordingly, consistent responses to SSD events that prevent any one SSD event from excessively depleting seed reserves may be adaptive in environments with frequent disturbance.

This study was motivated by several questions regarding emergence behaviour in frequently disturbed environments. These questions included: (i) does SSD promotion of seedling emergence always increase the population density of flushes rather than increase flush frequency?, (ii) can dissipation of SSD effects on emergence be quantified and incorporated into predictive models?, and (iii) does adaptation to local pedo-climatic conditions increase consistency in emergence behaviour, such that a single SSD event does not cause disproportionately large effects on emergence dynamics? To answer these questions, we quantified *Chenopodium album* L. emergence behaviour following SSD for local and common populations at 16 burial site-years located across Europe and North America. *Chenopodium album*, a common summer annual weed, is ideally suited for studying population and environmental factors influencing SSD-induced emergence, because it features photoblastic seeds that complete germination in response to SSD (Gallagher & Cardina, 1998), has differing emergence behaviours between geographically isolated populations (Eslami, 2011) and exhibits extended periods of emergence that facilitate within site-year comparisons (Mulugeta & Stoltenberg, 1997). Previous research indicated seasonality in the *C. album* emergence response to SSD, with more emergence following spring SSD compared with summer SSD (Roberts & Potter, 1980). Building on previous research, we hypothesised that: (i) regardless of burial site and seed population, SSD near the time of spring emergence universally promotes *C. album* seedling recruitment relative to undisturbed ground, by increasing recruitment from a given set of flushes rather than increasing the

number of flushes, (ii) the promotional effects of SSD on *C. album* emergence decrease with increasing time elapsed between SSD and flush occurrence and (iii) compared with a non-adapted population, locally adapted populations exhibit reduced recruitment variability within sites and across SSD events occurring at different times.

Materials and methods

Experimental approach and data collection

Emergence behaviour responses to SSD were studied in 16 site-years during 2008 and 2009 at 10 research farms in Europe and North America (Table 1). Seed populations were collected and buried during autumn 2007. At six study locations, experimental units that were established in 2007 were used for the collection of data in 2008 and 2009, with SSD treatments implemented on the same experimental units both years. At four study locations, emergence data were collected in 2008 only.

Prior to burial, seed viability was approximated by taking the higher of the results of germinating seeds at 10°/20°C 12 h/12 h and 5°/25°C 8 h/16 h fluctuating temperature regimes. Germination was tested in 9-cm-diameter polystyrene Petri dishes lined with two circles of Whatman No. 181 seed testing paper moistened with 4.5 mL of 0.01 M potassium nitrate solution (Analytical reagent grade; BDH Chemicals) prepared with deionised water. Each test comprised four replicates of 50 seeds. Dishes were placed in clear polythene bags and exposed to diffuse laboratory light. Germination criterion was 2 mm radicle emergence. Viability of the seed populations before burial was estimated to be high (>70%) in all except the Swedish seed population, where it was only 28.5%. However, the apparent low viability of the Swedish population may have been due to high levels of dormancy.

At each location, factorial combinations of two *C. album* seed populations and six SSD timing treatments were arranged in a randomised complete block design with two replications. Experimental units ('seed-banks') were aluminium mesh (1.5 mm opening) trays (25 cm width, 25 cm length, 5 cm depth) that were buried outdoors to a depth of 4 cm and filled with substrate mixed with 1000 seeds. The substrate consisted of soil, unfertilised peat and sand (2:1:1), with soil locally obtained and passed through a 9-mm sieve prior to use. To each cubic metre of substrate was added 0.6 kg ground limestone and 1.2 kg superphosphate. This recipe was based on a recipe of 'John Innes seed compost' (John Innes Institute, Norwich, UK, gardeningdata.co.uk) and was selected to provide as

Table 1 Burial site locations and dates and superficial soil disturbance (SSD) schedules

Site	Latitude	Longitude	Date of burial 2007	2008 SSD treatment timing*					2009 SSD treatment timing				
				SSD ₀	SSD ₅₀	SSD ₁₀₀	SSD ₁₅₀	SSD ₂₀₀	SSD ₀	SSD ₅₀	SSD ₁₀₀	SSD ₁₅₀	SSD ₂₀₀
České Budejovice, Czech Republic	48°58'N	14°27'E	31 October	12 March	29 March	5 April	19 April	1 May	9 April	17 April	24 April	3 May	13 May
Slagelse, Denmark	55°19'N	11°23'E	4 December	18 February	14 March	8 April	24 April	1 May	13 April	—†	23 April	29 April	12 May
Jokioinen, Finland	60°48'N	23°28'E	26 October	28 April	3 May	9 May	21 May	30 May	15 May	22 May	27 May	1 June	8 June
Urbana, IL, USA	40°20'N	88°14'W	1 November	29 March	8 April	17 April	23 April	26 April	17 March	25 March	3 April	17 April	24 April
Perugia, Italy	42°57'N	12°22'E	7 November	18 February	1 March	10 March	17 March	31 March	9 March	17 March	30 March	3 April	8 April
Morris, MN, USA	45°41'N	95°48'W	11 November	21 April	5 May	9 May	16 May	21 May	23 April	30 April	7 May	12 May	18 May
Ås, Norway	59°40'N	10°46'E	26 October	21 April	30 April	6 May	13 May	22 May	—	—	—	—	—
Zaragoza, Spain	41°43'N	0°48'W	30 November	4 March	14 March	18 March	25 March	1 April	—	—	—	—	—
Uppsala, Sweden	59°50'N	17°47'E	31 October	24 April	30 April	8 May	16 May	21 May	—	—	—	—	—
Warwick, UK	52°13'N	1°30'W	2 November	19 April	29 April	3 May	9 May	14 May	—	—	—	—	—

*SSD treatments were implemented based on time since first emergence, with time measured in day-degrees (d°C) calculated using a base temperature of 3°C. SSD treatments were as follows: undisturbed and 0, 50, 100, 150, 200 d°C after first emergence.

†SSD₅₀ did not take place at Slagelse, Denmark in 2009.

uniform a substrate across locations as possible, with the substrate similar to arable soil with regard to pH and phosphate content. Downwards seed losses were minimised with sheets of water-permeable, non-woven spunweb (Lutrasil®; Freudenberg Non-wovens, Weinheim, Germany or similar material) placed in the bottom of the trays. Damage from surface-foraging predators was prevented with mesh nets (≤ 9 mm openings).

Two seed populations were examined. 'Local' populations were matured on plants in agricultural fields near burial sites. The 'common' population, which was studied at all burial sites, matured on plants in an agricultural field at Aarhus University, Research Centre Flakkebjerg, Slagelse, Denmark (55°24'N 11°21'E). Research Centre Flakkebjerg was also the maturation location for the Denmark local population; however, the local population and the common population matured in different fields and were collected at different times. To harvest seeds, plants with mature seeds were shaken in a paper bag. Collected material was dried at 20–25°C in the absence of direct sunlight. Chaff was removed with combinations of sieving and forced-air separation. Seeds were then stored in moisture-proof containers at 3–5°C until used.

Seedbanks were monitored for emergence weekly during seasons conducive to *C. album* emergence. Emerged seedlings were counted and immediately removed by clipping hypocotyls at the soil surface, or by removing entire seedlings carefully with forceps without soil disturbance. The spring day on which emergence was first observed from any seedbank at a particular site initiated local scheduling of SSD treatments, which occurred at 0, 50, 100, 150, 200 d°C (day-degrees) after first emergence. Time after first emergence was calculated using daily mean air temperature data obtained from on-farm weather stations and a base temperature of 3°C, which approximated that for *C. album* seedling emergence in the literature (Harvey & Forcella, 1993; Vleeshouwers & Bouwmeester, 2001; Grundy *et al.*, 2003; Leblanc *et al.*, 2003; Gardarin *et al.*, 2010; Masin *et al.*, 2010). SSD was obtained by hand-mixing the substrate for several minutes in large, plastic containers. Hand-mixing was performed regardless of field moisture conditions, which facilitated precise scheduling of SSD events according to targeted day-degree intervals. Seedbanks that were not subjected to SSD ('undisturbed') served as controls for SSD.

We also measured the effects of soil moisture content at the time of SSD on SSD-induced emergence. Specific predictions regarding the effects of soil moisture content on SSD-induced emergence were

not made; rather soil moisture at time of SSD was considered a possible factor influencing variability in SSD-induced emergence. Gravimetric water contents of soil at times of SSD were determined with six extra trays, with one extra tray corresponding to each SSD event. SSD was applied to extra trays with hand mixing as described above. Immediately after SSD, a probe was used to extract a cylinder of soil (2 cm diameter, 5 cm height) that was weighed, dried at 100°C for 24 h and weighed again.

Statistical analyses

Overview

All statistical analyses were performed using the open source statistical software program R (v.3.0.1, The R Foundation for Statistical Computing, <http://www.r-project.org>). Emergence behaviour was summarised by measurements of seedling emergence over 1 year. Analytical units within this project included seedling emergence flushes, which were defined as consecutive weeks of gradually increasing then decreasing emergence. Maximum weekly emergence per counts flush were used to describe flush magnitudes. Flush magnitude means and maximums across 1 year, flush frequencies (flushes year⁻¹) and total emergence (seedlings year⁻¹) were measurements of annual emergence sequences.

Hypothesis 1: SSD promotes seedling recruitment relative to undisturbed ground by increasing recruitment per flush rather than number of flushes

Responses of individual emergence parameters to variable SSD timing were modelled using generalised linear mixed-effects models (GLMMs) and linear mixed-effects models (LMMs) developed with the R package *lme4*. In these models, SSD treatments were fixed effects. Random effects included replicate within site, site, population and all interactions among site, seed population and SSD treatment. Data for the 2 years of the study were analysed separately. For study locations in which trials were conducted in two consecutive years, total emergence for individual seedbanks in 2008 was not correlated with total emergence in 2009 ($r = 0.04$, $P = 0.58$, $n = 144$), thus indicating that annual runs represented independent trials that differed with respect to age of buried seed populations.

Disturbance effects on individual emergence parameters were evaluated with two statistical models. One model treated soil disturbance as a categorical variable and compared undisturbed seedbanks against all seedbanks subjected to SSD. A second model quantified emergence parameter responses to increasing d°C of

SSD treatment, a continuous variable. Models for total emergence responses to the bivariate predictor 'disturbance' and models for flush frequency responses to categorical and continuous variables for disturbance were GLMMs fitted using Poisson distributions and logarithms as link functions. Lack of model convergence prevented use of GLMMs for total emergence responses to increasing d°C of SSD treatment and instead, this relationship was modelled using LMMs following log-transformations of the dependent variable. LMMs with log-transformed dependent variables were also used to quantify relationships between flush magnitude and soil disturbance treated as both categorical and continuous variables.

Resulting GLMMs and LMMs included parameter estimates for intercepts and treatments. Intercept estimates corresponded to the means for undisturbed seedbanks, whereas means for treatment estimates differed by type of predictor variable. For the categorical predictor variable (bivariate of 'disturbance'), treatment estimates represented the differences between undisturbed seedbanks and seedbanks subjected to SSD. For the continuous predictor variable (d°C of SSD), treatment estimates corresponded to the slopes of graphs of emergence parameters against d°C of SSD. Model estimate standard errors were used to determine 95% confidence intervals (Pinheiro & Bates, 2000). Confidence intervals (95%) that included zero were indicative of non-significant effects on emergence parameters (Crawley, 2007).

Hypothesis 2: Promotional effects of SSD on emergence decrease with increasing time elapsed between SSD and flush occurrence

The relationship between the promotional effect of SSD on a particular flush and the time elapsed from disturbance was determined by first quantifying differences between SSD and undisturbed seedbanks in weekly emergence counts:

$$\Delta EMG_{nri} = SSD_{nri} - UD_{ri} \quad (1)$$

where ΔEMG_{nri} is the change in emergence caused by SSD timing treatment n in replicate r at day i ; SSD_{nri} is the weekly count of emerged seedlings from a seedbank subjected to disturbance timing treatment n in replicate r at day i ; UD_{ri} is the weekly count of emerged seedlings from an undisturbed seedbank in replicate r at day i . Days (i) were measured in d°C (base temperature of 3°C) accumulated from the day of disturbance. Values for ΔEMG_{nri} greater than zero were converted to proportions of the yearly total difference in emergence between a specific SSD seedbank and the corresponding undisturbed seedbank:

$$\begin{aligned} &\text{if } \Delta\text{EMG}_{nri} > 0, \text{ then} \\ &\tau_{nri} = \frac{\Delta\text{EMG}_{nri}}{\Phi\text{EMG}_{nr}} \end{aligned} \quad (2)$$

where τ_{nri} is the proportionate strength of SSD effect on emergence for disturbance timing treatment n in replicate r at day i , and ΦEMG_{nr} is the summation of ΔEMG_{nri} values greater than zero over 1 year.

Values for τ_{nri} were plotted as functions of time elapsed since disturbance, which is hereafter referred to as the ‘disturbance lag’. The impacts of disturbance lags on SSD promotion of emergence was quantified with a GLMM developed with the R package *lme4*. In this model, disturbance lag was treated as a fixed effect. Random effects included year, site, replicate within site, population, SSD treatment and all interactions among year, site, seed population and SSD treatment. Dependent variables of the GLMM (τ_{nri}) were binary response variables that were created from ΔEMG_{nri} values greater than zero. Specifically, for each ΔEMG_{nri} value greater than zero, a new data set was made with the number of entries for these data sets equal to the corresponding ΦEMG_{nr} value. Data set entries were then coded as ‘1’ for promotion of emergence, ‘0’ for no effect on emergence, with the number of 1s equal to ΔEMG_{nri} . The GLMM was fitted using a binomial distribution and a logit-link function. Significance of GLMM parameter estimates was evaluated with Wald tests using Z-statistics (Agresti, 2002). The exponentiated value of the model parameter estimate for disturbance lag provided the odds ratio (ratio of the odds of promotion of emergence to the odds of no promotion of emergence), which described the change in promotion of emergence when disturbance lag increased by one d°C (Agresti, 2002).

Hypothesis 3: Compared with a non-adapted population, locally adapted populations exhibit reduced recruitment variability within sites and across SSD events

Adaptive emergence behaviour in SSD environments was clarified by comparing variances. These variances were calculated for total emergence for each seed population within a specific site. Data for undisturbed seedbanks were not included in this analysis. Therefore, variances represented measures of spread in total emergence across SSD treatments and replicates within a specific site. Seed population effects on variance in total emergence were determined with a LMM (fitted with the R package *lme4*) that treated population type (local, common) as fixed effects. Random effects included year, site and interactions between population and year, population and site, and year and site. Random effects did not include a three-way interaction between population,

year and site because of an insufficient number of observations. To meet the assumptions of constant variance, data for total emergence were square-root-transformed prior to analysis. Resulting LMMs included parameter estimates intercept and treatment. The intercept estimate corresponded to the mean for the common population, whereas the treatment estimate represented the difference between the common and local populations. Model estimate standard errors were used to determine 95% confidence intervals (Pinheiro & Bates, 2000). The statistical significance of population effects on variance in total emergence was determined with 95% confidence intervals determined from standard errors of fixed-effect parameter estimates (Crawley, 2007).

Effects of soil moisture content at time of SSD on SSD-induced emergence

Relationships between soil moisture at the time of SSD and SSD-induced emergence were assessed through a two-step process. First, for each seedbank subjected to SSD, total annual emergence was divided by total annual emergence of the undisturbed seedbank corresponding with the same site, population and replicate. Hereafter, total annual emergence in SSD seedbanks divided by total annual emergence in undisturbed seedbanks is referred to as ‘relative emergence’. Relative emergence values were regressed against values for gravimetric water contents of soil at time of SSD. To these data, a linear regression model was fitted. Visual inspections of residuals plotted against fitted values indicated that variance systematically increased for larger predicted values of relative emergence. Such violations of assumptions of homoscedasticity were resolved by ln-transformations.

Results

Summary of emergence behaviour

Annual emergence sequences were often characterised by an initial high-density flush followed by several smaller flushes. However, this emergence pattern was not universal, as some emergence sequences were successions of flushes with similar densities (Fig. 1, Appendix S1). Burial environment factors that contributed to variability in emergence behaviour following SSD included soil moisture at the time of SSD, which was negatively related to the stimulatory effect of SSD on emergence (Fig. 2).

Hypothesis 1: SSD increases recruitment per flush

Compared with undisturbed soil, SSD increased the number of seedlings that emerged over 1 year (Table 2).

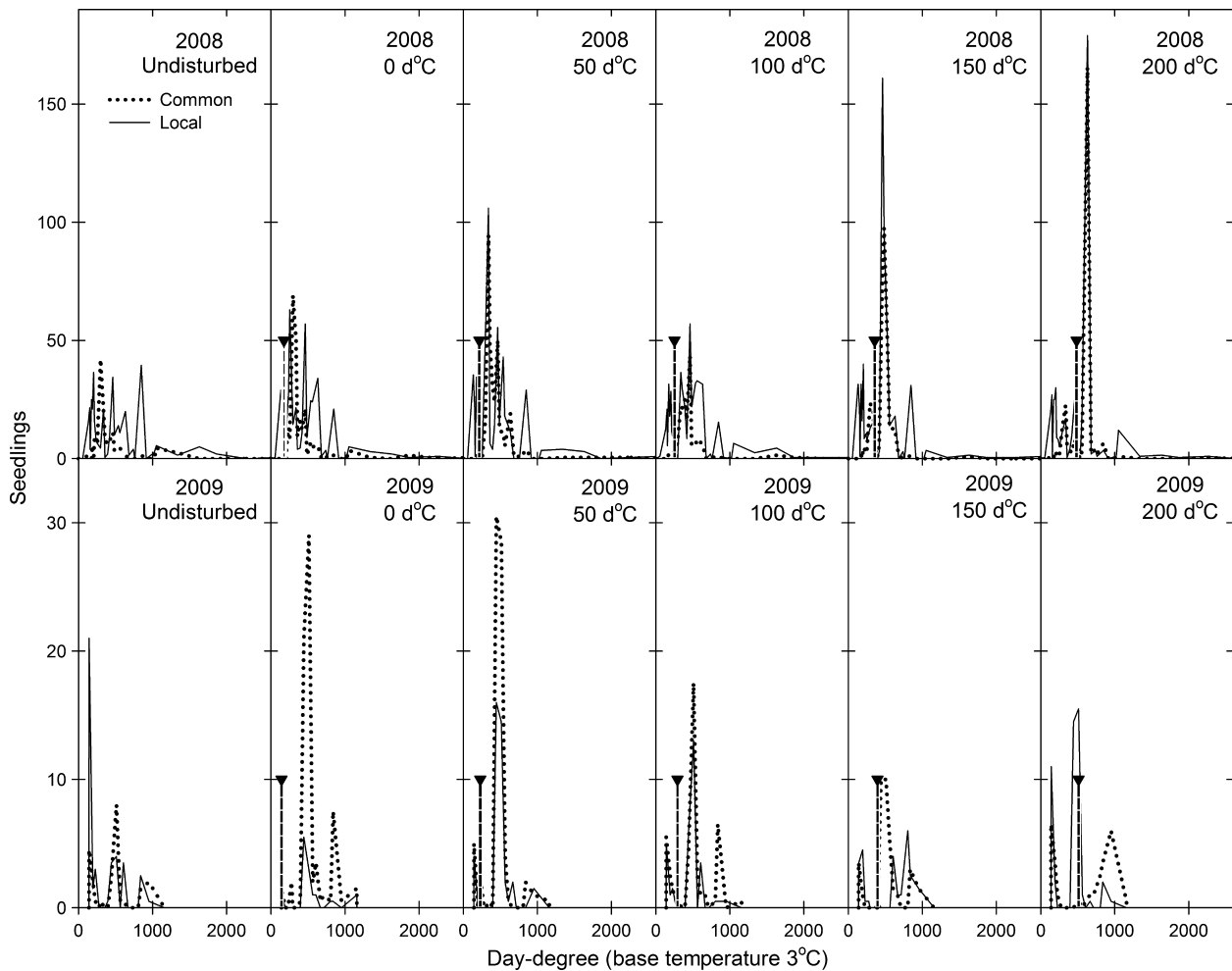


Fig. 1 Annual emergence sequences during 2008 and 2009 for common (dotted line) and local (solid line) seed populations buried in artificial seedbanks (25 cm by 25 cm, 5 cm depth) at České Budějovice, Czech Republic. Panels present superficial soil disturbance (SSD) treatments occurring at 0, 50, 100, 150 and 200 day-degrees (d°C; base temperature, 3°C) after first emergence from undisturbed soil. X-axis d°C accumulated from date of seedbank establishment (2008) and 1 year after this date (2009). Dashed lines indicate when SSD occurred. The common lot was studied at all burial sites (full emergence profiles are provided in Appendix S1).

One year after burial (2008), the increase in total emergence was not associated with increases in flush frequency but coincided with greater flush magnitudes. Mean flush magnitude in 2008 was 1.8 times greater for SSD seedbanks than for undisturbed soil, and maximum flush magnitude was 2.2 times greater for SSD seedbanks. Two years after burial (2009), seedbanks subjected to SSD exhibited greater total emergence and greater flush frequency relative to undisturbed seedbanks. SSD seedbanks in 2009 also featured greater maximum and mean flush magnitudes. The increase in total emergence in 2009 that was caused by SSD (43 seedlings) was greater than the increase in mean flush magnitude caused by SSD (five seedlings). This indicates that the increase in total emergence in 2009 that was caused by SSD was not exclusively attributable to the increase in flush frequency. Thus, 2 years after burial, SSD impacted emergence behaviour by increasing

total emergence through, at least in part, increased flush magnitude.

The timing of SSD relative to first emergence influenced its promotional effects on emergence in 2008, but not in 2009 (Table 3). Specifically, delayed SSD in 2008 increased total emergence, mean flush magnitude and maximum flush magnitude, whereas delayed SSD in 2009 did not affect emergence parameters.

Hypothesis 2: Promotional effects of SSD decrease over time

The proportionate strengths of SSD promotional effects on emergence (τ_{mri} ; Eqn 2) were negatively related to the time elapsed between that flush and a particular SSD event (i.e. the disturbance lag; Fig. 3). Examples of the negative association between disturbance lag and SSD promotional effects included sev-

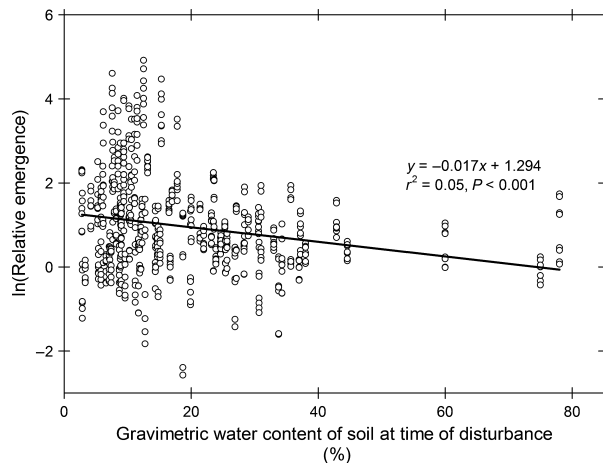


Fig. 2 The response of ln-transformed relative emergence to increasing soil moisture at time of superficial soil disturbance (SSD). Relative emergence is total annual emergence in SSD seedbanks divided by total annual emergence in undisturbed seedbanks.

eral seedbanks for which all SSD-induced emergence occurred in a single emergence event taking place within 500 d°C of disturbance. The negative association between disturbance lag and proportionate strength of SSD promotion of emergence was statistically significant, as indicated by a statistical model that revealed diminishing SSD promotion of emergence by 0.21% with each elapsed d°C between SSD and a particular seedling flush (Table 4). Notwithstanding the negative association between proportionate strengths of SSD promotional effect and disturbance lag, low levels of SSD-induced emergence were detected as late as 3000 d°C after disturbance [e.g. in Perugia (Appendix S1)].

Hypothesis 3: Locally adapted populations exhibit reduced recruitment variability

For seedbanks subjected to SSD, total annual emergence was characterised by high levels of variation within sites and across replicates. For seed populations of specific site-years, coefficients of variation for total emergence were as great as 136%. Variance was significantly affected by population, with greater variance in total emergence for the common population than for the local populations (Table 5).

Discussion

The goals of this study were to quantitatively test assumptions on SSD-induced emergence and to offer perspectives on adaptive emergence behaviours in environments characterised by frequent SSD. Before providing context to the results, we reiterate that SSD

Table 2 Parameter estimates (β), with 95% confidence limits, from generalised linear mixed models of soil disturbance effects on total emergence, flush frequency and flush magnitude. Models were fitted to data for *Chenopodium album* seedling emergence data from 10 sites in 2008, six sites in 2009. Data for 2008 pertain to seed populations buried 1 year, seed populations were buried for 2 years in 2009

Year	Parameter*	Total emergence		Flush frequency		Maximum flush magnitude		Mean flush magnitude	
		β †	Seedlings year ⁻¹	β	Flush year ⁻¹	β	Seedlings per flush	β	Seedlings per flush
2008	Intercept (undisturbed)	4.09 (3.560; 4.628)	60	1.42 (1.333; 1.761)	4.7	2.87 (2.264; 3.467)	18	2.24 (1.737; 2.744)	9
	Disturbed	0.78 (0.523; 1.028)	130	0.05 (−0.103; 0.203)	4.9	0.83 (0.514; 1.155)	40	0.56 (0.331; 0.786)	16
2009	Intercept ((undisturbed)	2.40 (1.806; 2.996)	11	1.04 (0.659; 1.423)	2.8	1.82 (1.191; 2.443)	6	1.26 (0.906; 1.618)	7
	Disturbed	1.58 (0.884; 2.737)	54	0.29 (0.042; 0.534)	3.8	1.04 (0.204; 1.879)	17	0.89 (0.423; 1.361)	12

*Seedbanks were binomially classified as 'disturbed' or 'undisturbed', with undisturbed seedbanks coded as the reference category in the generalised linear mixed model.

†Parameter estimates are presented in the linked scale, followed by the back-transformed mean predicted by the model. Models for total emergence and flush frequency were fitted using Poisson distributions, whereas models for flush magnitude were fitted using Gaussian distributions following log-transformations of the dependent variable. Parameter estimates for intercepts correspond to the means for undisturbed seedbanks. Estimates for the parameter 'disturbed' represent the differences between disturbed and undisturbed seedbanks.

Table 3 Parameter estimates (β), with 95% confidence limits, from generalised linear mixed models of soil disturbance timing effects on total emergence, flush frequency and flush magnitude. Models were fitted to data for *Chenopodium album* seedling emergence data from 10 sites in 2008, six sites in 2009. Data for 2008 pertain to seed populations buried 1 year, whereas seed populations were buried 2 years in 2009

Response	Predictor*	β † (95% confidence limits)	
		2008	2009
Total emergence	Intercept	4.59 (4.048; 5.147)	3.90 (3.283; 4.516)
	Disturbance treatment d°C	1.9×10^{-3} (1.0×10^{-3} ; 2.7×10^{-3})	-1.6×10^{-3} (-3.1×10^{-3} ; 4.5×10^{-5})
Flush frequency	Intercept	1.60 (1.411; 1.786)	1.45 (1.118; 1.775)
	Disturbance treatment d°C	1.9×10^{-4} (-6.7×10^{-4} ; 1.0×10^{-3})	-1.1×10^{-3} (-2.4×10^{-3} ; 1.1×10^{-4})
Maximum flush magnitude	Intercept	3.44 (2.889; 3.996)	3.02 (2.425; 3.619)
	Disturbance treatment d°C	2.6×10^{-3} (1.4×10^{-3} ; 3.8×10^{-3})	-1.3×10^{-3} (-3.2×10^{-3} ; 5.0×10^{-4})
Mean flush magnitude	Intercept	2.66 (2.176; 3.159)	2.16 (1.775; 3.619)
	Disturbance treatment d°C	1.3×10^{-3} (3.9×10^{-4} ; 2.2×10^{-3})	-6.3×10^{-5} (-1.4×10^{-3} ; 1.3×10^{-3})

*Soil disturbance treatments occurred at 0, 50, 100, 150 and 200 d°C (base temperature, 3°C) after first emergence from undisturbed soil. Day-degrees of soil disturbance were modelled as continuous variables, and thus, parameters for disturbance treatment represent the impacts of d°C increases on respective emergence parameters.

†Models for flush frequency were fitted using Poisson distributions, whereas models for total emergence and flush magnitude were fitted using Gaussian distributions following log-transformations of the dependent variable.

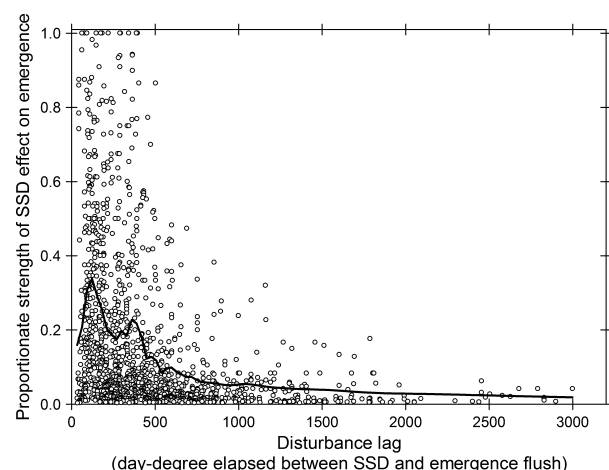


Fig. 3 The relationship between proportionate strengths of superficial soil disturbance (SSD) promotional effects on emergence (τ_{nr} ; Eqn 2) and the time elapsed, in day-degrees (d°C), between a flush and a particular SSD event. Strengths of SSD promotional effects are proportions of the total SSD effect on emergence for a given SSD treatment replicate at a specific site-year. The solid line presents a locally weighted regression model.

was implemented through hand-mixing, which may have caused greater soil disturbance and increased the proportion of seeds stimulated to germinate by light,

compared with SSD more typical of practical cropping situations. Accordingly, flush magnitudes following SSD reported here may be high compared with flush magnitudes following SSD in cropping situations. Despite this methodological limitation created by the need for precision in SSD timing, our results confirmed and expanded previous observations, such that a robust framework for understanding SSD-induced seedling emergence should include the following principles: (i) 1 year after burial, SSD promotes emergence by increasing densities of seedling flushes rather than increasing flush frequency; (ii) the promotional effects of SSD on seedling flush population density dissipate over time; and (iii) local adaptation in emergence behaviour entails increased consistency in emergence responses to SSD events at different times.

Unexpectedly, we discovered that promotional effects of SSD on emergence were conditioned by the age of the buried seed population. These results were consistent with those of Ogg and Dawson (1984), who showed that for many weed species, including *C. album*, temporal patterns of emergence over a 1-year period differed between seed populations buried 1 or 2 years. Because weed seedbanks are comprised of seeds that differ in age, neglecting the potential

Table 4 Summary of a logistic regression model that quantified the effects of increasing disturbance lag [i.e. time elapsed between a seedling flush and a superficial soil disturbance (SSD) event] on the promotional effects of SSD on seedling emergence

Parameter	β	SE β	Z-statistic*	P	e^{β} †
Intercept	-1.03	0.016	-62.3	<0.001	
Disturbance lag	-2.21×10^{-3}	3.08×10^{-5}	-69.6	<0.001	0.998

*Z-statistic for maximum likelihood estimate, which is equal to $(\beta/\text{SE } \beta)$, where β is the maximum likelihood parameter estimate.

†Exponentiated value of the parameter estimate for disturbance lag, which describes the change in probability in promotion of emergence when disturbance lag increased by one unit. Disturbance lags were measured in day-degrees, base temperature 3°C.

Table 5 Summary of a linear mixed-effect model that quantified seed population effects on variance in total emergence among disturbed seedbanks within site-years. The model was fitted following a square-root-transformation of the dependent variable

Parameter*	β	95% confidence limits for β
Intercept (Common population)	38.49	(5.095; 71.891)
Local population	−6.86	(−13.623; −0.097)

*Each site-year contained two seed populations (local and common, with the common population studied at all site-years). The parameter estimate (β) for intercept corresponds to the mean for the common population, whereas the parameter 'local population' represents the difference between local and common populations.

impacts of burial duration on emergence behaviour can lead to erroneous predictions of emergence patterns. Thus, predictive models for weed seedling emergence may be improved with increased knowledge of the mechanisms underlying yearly differences in emergence behaviour for a specific seed population.

We discovered that SSD impacts on emergence behaviour were not exclusively confined to narrow temporal windows, because low levels of SSD-induced emergence occurred as late as 3000 d°C after SSD. In general, sensitivity to triggers of germination is determined by progressions in dormancy loss and secondary dormancy induction (Benech-Arnold *et al.*, 2000). These processes are regulated by microenvironment and genetic factors that vary between individual seeds such that, at any given time, buried seed populations contain a range of sensitivities to germination triggers. Intrapopulation variation in germination sensitivity is evidenced by results from population-level, bury-and-recover studies that rarely show 100% germination at any specific time (e.g. Schutte *et al.*, 2012). Our results suggest SSD creates an effect that eventually promotes germination of highly dormant seeds incapable of immediate responses to SSD. This delay in SSD-induced emergence is perhaps caused by seed movement to soil microenvironments that become more conducive to either germination or seedling emergence, compared with the original burial locations. Previous research has shown that the vertical distribution of weed seeds in soil, which can influence dormancy loss (Omami *et al.*, 1999) and emergence likelihood (Harrison *et al.*, 2007), is affected by soil disturbances (Mohler *et al.*, 2006).

A framework for understanding SSD-induced emergence broadly benefits understanding of weed infestation parameters that are intrinsically tied to the magnitudes and timings of seedling flushes (i.e. interference potentials and population dynamics). Such

information is important for agricultural approaches that emphasise weed management through exploitation of biological and ecological processes. Management implications stemming from this study include several points of guidance on *C. album* seedbank depletion through stale seedbed methods. First, because SSD-induced emergence was influenced by both seed population and burial site factors, recommendations for scheduling stale seedbed disturbances should remain local. Local factors that influence SSD-induced emergence include soil moisture percentage at the time of SSD (Fig. 2). Therefore, recommendations for stale seedbed disturbances should account for interactions between SSD and soil moisture that impact emergence behaviour following SSD. Despite the importance of local conditions on SSD-induced emergence, our results suggest a general timeframe for initiating stale seedbed disturbances applicable across broad spatio-temporal zones. This is because SSD treatments occurring at 0, 50, 100, 150 and 200 d°C after first emergence each promoted emergence (Table 2). However, any timeframe for SSD scheduling needs to be confirmed with additional studies that subject seedbanks to repeated SSD events within a single growing season.

Additional guidelines for stale seedbed practices resulting from this study pertain to the timing of control for emerged weed seedlings. Because SSD effects on emergence were reduced after *c.* 500 d°C following SSD, extended intervals between SSD and weed control may not improve stale seedbed effectiveness towards weed seedbank depletion. Finally, because of the potential for prolonged delays in SSD-induced emergence, implementation of stale seedbed tactics should be accompanied with anticipation of minor post-disturbance increases in weed densities by the end of the growing season.

Few studies have focused on relationships between soil moisture content and the stimulatory effect of SSD on emergence. In our study, the negative relationship between SSD-induced emergence and soil moisture percentage at the time of SSD may have been caused by increased inhibition of seedling emergence by soil compaction. This is because (i) soil compaction is generally detrimental to seedling emergence (e.g. Hyatt *et al.*, 2007) and (ii) soil compaction after SSD becomes more severe with increasing soil moisture content at time of SSD (Hamza & Anderson, 2005). Despite the possible influence of soil compaction on seedling emergence, the relationship between soil moisture percentage at the time of SSD and SSD-induced emergence was weak, as evidenced by the low r^2 for a linear model for relative emergence responses to increasing gravimetric water content of soil at time of

SSD (Fig. 2). The simple regression for the influence of soil moisture percentage at the time of SSD on SSD-induced emergence did not account for the stochastic factors that influence emergence behaviour following SSD, most notably, subsequent precipitation patterns.

Superficial soil disturbance-induced emergence for *C. album* occurred after light-induced germination (Gallagher & Cardina, 1998). Previous studies have clarified impacts of moisture content of the incubation medium on germination responses of light-exposed seeds (Gallagher & Cardina, 1997; Botto *et al.*, 2000). These studies indicated germination responses of seeds exposed to light are inhibited by low soil moisture (<10% volumetric soil moisture content; Gallagher & Cardina, 1997). Results of previous studies combined with data from our investigation suggest that, for a population of photoblastic seeds, SSD-induced emergence displays a unimodal response to increasing soil moisture content, with maximum SSD-induced emergence occurring at moderate soil moisture levels. This hypothesised response could be tested by applying SSD to seedbanks held at a different levels evenly distributed across a wide soil moisture gradient.

Variability in a specific trait is confined by bounds established by an organism's life history evolution, which for *C. album* has led to rapid emergence following disturbance. Genetic factors that regulate germination in response to disturbance may be among the collection of traits conserved across *C. album* populations in agricultural environments, thereby facilitating conservation of its ecological niche. Although emergence in response to disturbance may be a conserved trait in *C. album*, results of this study suggest *C. album* populations differ in abilities to 'bet hedge', that is, to reduce temporal variation in fitness by sampling a range of recruitment environments through time (Childs *et al.*, 2010). Bet-hedging strategies are potentially evolved traits (Clements *et al.*, 2004) and, as such, the degree of offspring diversification is likely to differ between individuals and populations with divergent evolutionary histories. Evolved abilities to 'bet hedge' are consistent with *C. album*'s presence in a broad range of cropping systems and emphasise the importance of preventive management tactics to limit *C. album* plant establishment. In addition to these insights into *C. album* evolution, the results of this study advance a robust framework for understanding SSD-induced weed seedling emergence, which is a common occurrence in managed crop fields, a central component of weed seedbank depletion, and a mainstay of non-chemical weed management. How other weeds species respond in this respect need to be investigated in other studies.

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Supporting Information

Additional Supporting Information may be found in the online version of this article:

Appendix S1 Annual emergence sequences for common (dotted line) and local (solid line) seed lots buried at Slagelse, Denmark; Jokioinen, Finland; Urbana, IL, United States; Perugia, Italy; Morris, MN, United States; Ås, Norway; Zaragoza, Spain; Uppsala, Sweden; and Warwick, United Kingdom.